Instructions for Use

Leitz

Phase Contrast Equipment

with the Heine Condenser

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The LEITZ Phase Contrast Equipment permits the setting of the following types of microscope illumination:

Brightfield Illumination
Phase Contrast after Zernike
Darkfield Illumination

The Heine condenser used in the equipment renders it possible to set these kinds of illumination while observing the specimen, by operating a control knob on the condenser, and thus to use every intermediate adjustment in continuous transition from brightfield to phase contrast or to darkfield.

The possibility thus offered of continually changing the illumination is of particular advantage in cases where the structures visible in phase contrast are to be compared with the structures visible in an ordinary brightfield or a darkfield. According to the particular nature of the specimen, the intermediate settings can also be important for differentiating, and can assist in the judgement of the specimen.

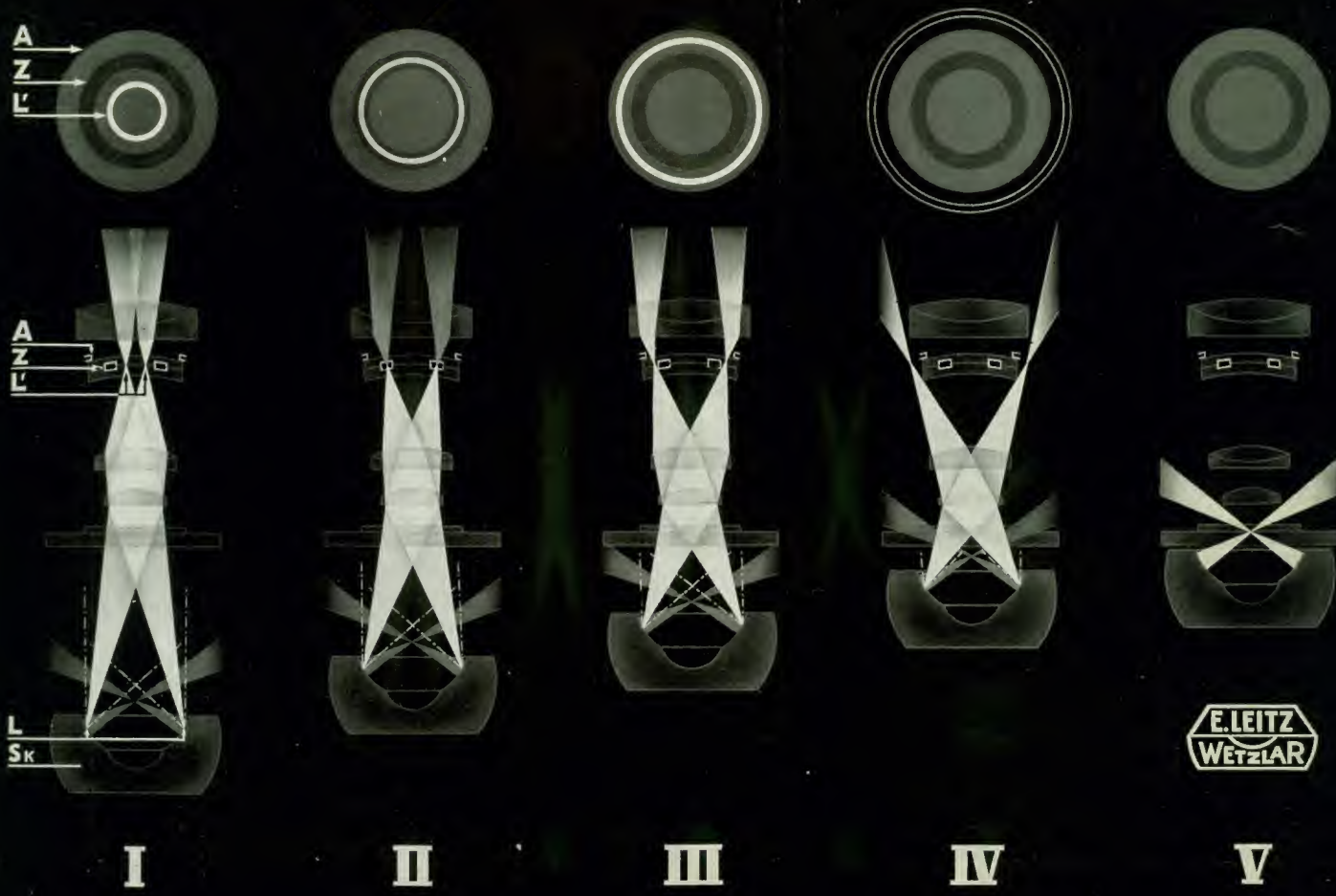
The LEITZ Phase Contrast Equipment

consists of: the Heine condenser (1) with control knob (Tr) for the vertical adjustment of the built-in mirror component (Sk), immersion cap (1 a) to screw to the condenser, a set of objectives with suitably selected apertures, i. e., achromatic dry system Pv 10/0.25*) (2), immersion attachment (2 a) for the objective Pv 10/0.25, achromatic dry system Pv 20/0.45 (3), apochromatic dry system Pv Apo 40/0.70 in correction mount with automatic focusing compensation (4), apochromatic oil immersion objective Pv Apo Oil 90/1.15 (5), filter holder (6) with daylight and photographic filters (6 a), auxiliary magnifier (7).

The equipment can be supplied to fit any LEITZ microscope; for the ORTHOLUX and PANPHOT microscopes it is supplied with a dovetail slide fitting the horizontal condenser holder, for all other stands fitted with condenser sleeve (internal diameter of the sleeve: 39.5 mm.) with cylindrical sliding mount. The Heine condenser can also be used on the students' microscope H; in this case a special extension section for the mirror holder is required.

As the microscope lamp for stands without built-in sources of light, the MONLA low voltage lamp (6 volts 5 amps.) is to be recommended, preferably in the MONLAFIX design on which a special pamphlet is available.

*) The number before the oblique stroke gives the initial magnification, the figure after the stroke gives the numerical aperture.


A
Z
L'
A
Z
L'
L
SK
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I
II
III
IV
V

The various types of illumination

in relation to the position of the mirror component. Objective Pv 20/0.45.

With the mirror component Sk in the lowest position (I) the narrow ring of light L produced by the condenser is reduced to L' within the Zernike phase ring Z. Brightfield observation is given.

As the mirror component Sk is raised, the image L' of the ring of light widens until it is completely covered by the dark-looking phase ring Z. Position II has now been reached with phase contrast after Zernike.

Further raising of the mirror component Sk permits the image of the illumination ring to increase still further, until it is no longer influenced by the phase ring. This position (III) gives brightfield images with very rich contrast qualities.

Further raising of the mirror component Sk causes the image of the light ring to vanish beyond the edge A of the aperture diaphragm. In this position (IV) with the ring L as source of light, a particular darkfield is achieved which in many cases reveals special structures more clearly than ordinary darkfield.

Finally in position V the cone of illumination convergent in the object field becomes effective; this gives normal darkfield illumination.

The transition from one of the characteristic settings to the other is continuous.

The phenomena in the aperture diaphragm of the objective can be observed with the auxiliary magnifier, which is inserted into the body tube instead of the eyepiece (focusing on the light ring by turning the eyelens).

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The Objectives

All objectives supplied with the equipment are corrected for a tube length of 170 mm. and a cover glass thickness of 0.17 mm. Initial magnification and aperture are appropriately selected. If a microscope stand is available with interchangeable revolving nosepiece carrier (e. g., the stands B, ORTHOLUX, PANPHOT), it is advisable to order the phase contrast objectives par-focal on a special revolving nosepiece, and not to remove them or interchange them on this revolving nosepiece, in order to maintain the uniform centration.

The achromatic objective Pv 10/0.25 serves as a general survey objective. Even with the mirror component in the lowest position it already shows a phase contrast image, and later a darkfield image. An immersion attachment is provided for the objective Pv 10/0.25. It is used when working with the objective Pv Apo Oil 90/1.15. The objective Pv 10/0.25 then serves to seek out suitable sites of the specimen, the immersion attachment giving the same working distance as that for which objective Pv Apo Oil 90/1.15 is designed; it is thus possible to switch directly from one objective to the other.
The achromatic objective Pv 20 : 1 has the numerical aperture A = 0.45.

The apochromatic objective Pv Apo 40/0.70 has cover glass correction with automatic focusing compensation.

High quality dry objectives can only become fully effective with cover glass correction. But the ordinary cover glass correction has the disadvantage that on adjusting to the cover glass thickness the focusing is so greatly altered that the image is again completely lost, and on re-focusing it is easily possible to focus in the wrong direction at first. The automatic sharpness correction, on the other hand, completely avoids this disadvantage. The correction mount is set to the average cover glass thickness of 0.17 and the image is sharply focused; the best image quality is then sought by turning the knurled objective ring, and the fine focusing is easily subsequently achieved.

The mount of the oil immersion Pv Apo Oil 90/1.15 has been so lengthened that even the strong dry objective cannot come into contact with the oil drop on changing over. On the other hand, with careless changing over, the objective mount of the immersion objective strikes the specimen carrier prematurely.

The apochromatic oil immersion Pv Apo Oil 90/1.15 makes use of the highest available aperture of the Heine condenser in darkfield in position IV. In combination with our darkfield condenser D 1.20 A this apochromatic objective gives the best darkfield which can possibly be achieved.

Directions for Use

1. Adjust the light in the microscope most expediently without condenser, without objective and without eyepiece on looking into the body tube (with ORTHOLUX and PANPHOT the pivoting illuminating lens should be swung out of action). With microscopes with built-in sources of light, or when using the MONLAFIX microscope lamp, the centration of the source of light can be easily checked with the standard centred bright-field condenser.
2. Insert the Heine condenser and attach the set of objectives on the microscope. Operate the rack and pinion for the vertical adjustment of the substage upwards against the stop. This position remains unaltered during the examination. The mirror component Sk should be set in the lowest position against the stop in its cylindrical guide by means of the control knob Tr. Objective Pv 10/0.25 on the revolving nosepiece should then be swung into position. The eyepiece used is the periplanatic eyepiece 8 x.
3. With the mirror component in its lowest position, focus the image sharply with the coarse and fine focusing mechanism of the microscope.
4. Regulate the brilliance of the lamp; illuminate the field of view uniformly by focusing the illuminating lens on the microscope lamp.
5. Remove the eyepiece and insert the auxiliary magnifier in the body tube. Focus the eyelens sharply on the light ring L' and centre the latter to the aperture iris edge A by operating the two lateral screws S of the condenser. The centring is simplified if the mirror component is raised so high that the light ring just touches the diaphragm edge; as the mirror component is further raised, the light ring should vanish uniformly from the field of view. The mirror component is then returned to the lowest position by means of the control knob Tr. When centring, reduce the brilliance of the lamp by regulating the transformer if necessary. Replace the eyepiece.
6. If the objectives are not supplied par-focal on a special revolving nosepiece, a slight subsequent focusing may prove necessary when changing the objectives. The best centring is achieved if on raising the mirror component to its top position (by means of control knob Tr), a shadow spot becomes faintly visible in the centre. Slight subsequent centring to this shadow spot may be necessary.

7. Phase contrast is obtained with the objective Pv 10/0.25 with the mirror component in its lowest position; with the other objectives the mirror component must be set correspondingly higher.

8. When using the immersion objective Pv Apo Oil 90/1.15 the immersion cap is screwed onto the Heine condenser, given a drop of oil, and brought upwards to the object slide from beneath by means of the rack and pinion of the substage, until the oil drop comes into contact with the object slide. Take care that the object slide is not pushed up by the immersion cap. The condenser then remains in this position during the observation with immersion. The adjustment of the brightfield, phase contrast or darkfield image is effected by means of the mirror component control knob Tr (with objective Pv Apo Oil 90/1.15 without the immersion cap on the condenser, a phase contrast image can still be achieved, but neither a darkfield nor a brightfield image is possible).

   Before bringing the immersion objective into use, the body tube must be raised, or the stage must be lowered according to the type of microscope. The drop of oil applied to the specimen should not be too small, and the oil immersion should then be lowered until it dips into the oil drop (distance from cover glass 0.13 mm.). Focus the image sharply by means of the micrometer screw. If objective Pv 10/0.25 is used with the immersion attachment for locating suitable sites of the preparation, no adjustment to the coarse focusing is necessary on changing to oil immersion (see also page 6).

9. Although as a rule work is carried out without the pivoting illuminating lens on the ORTHOLUX and PANPHOT and without the front lens on the MONLAFIX, when setting normal darkfield (Position V) with the two low power objectives, it is advisable to bring these lenses into action in order to achieve uniform illumination of the image field.

10. The cover glass and the front lens of the objective must always be spotlessly clean. The eyelens should similarly be cleaned frequently, since with the small circular exit pupil the image is already affected through slight soiling by the eye-lashes.

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